AMENDMENTS TO THE CLAIMS (4/7/04)

1. (currently amended) A method for determining the concentration of chloride ions in samples, comprising:

combining an enzyme reagent with an α-amylase activity detecting substrate, a sodium ion and a sample containing a chloride ion to be assayed, wherein the enzyme reagent includes α-amylase that is substantially calcium-free and wherein the concentration of the sodium ion is at a level so that α-amylase is substantially activated by the sodium ion in proportion to the amount of the chloride ion in said sample ~~present in a concentration higher than the concentration of said chloride ion~~;

assaying the quantity of α-amylase activated by the sodium ion ~~in proportion to the amount of the chloride ion in said sample~~; and

determining the quantity of said chloride ion by reference to said activity of α-amylase.

2. (original) The method according to claim 1, wherein calcium is removed from the α-amylase that is substantially calcium-free by use of a chelating compound.

3. (original) The method according to claim 1, wherein calcium is removed from the α-amylase that is substantially calcium-free by use of a compound that forms a covalent bond with calcium.

4. (original) The method according to claim 2, wherein said chelating compound is a member selected from the group consisting of ethylenediaminetetraacetic acid, trans-1,2-cyclohexanediamine-N,N,N',N'-tetraacetic acid, glycol ether diamine tetraacetic acid, iminotetraacetic acid, and diaminopropanetetraacetic acid.

5. (original) The method of claim 2, wherein said chelating compound is ethylenediaminetetraacetic acid.

6. (original) The method according to claim 1, wherein said α-amylase activity detecting substrate is a member selected from the group consisting of 2-chloro-4-nitrophenyl-α-D-maltotrioside, 4-nitrophenyl-α-D-maltopentaoside and α-glucosidase, 2-chloro-4-nitrophenyl-β-D-maltopentaoside and α-glucosidase and β-glucosidase, 4-nitrophenyl-α-D-maltoheptaoside, α-glucosidase, and 2-chloro-4-nitrophenyl-β-D-maltoheptaoside and α-glucosidase and β-glucosidase.

7. (original) The method according to claim 6, wherein said α-amylase activity detecting substrate is 2-chloro-4-nitrophenyl-α-D-maltotrioside.

8. (original) The method according to claim 1, wherein said sample is a bodily fluid sample.

9. (previously presented) The method according to claim 8, wherein said bodily fluid sample is selected from the group consisting of serum, plasma, and urine.

10. (currently amended) The method of claim 1, wherein said sodium ion is in the form of sodium citrate.

11. (currently amended) The method of claim 1, wherein said sodium ion is in the form of sodium acetate.

12. (currently amended) A composition for use in determining the concentration of a chloride ion in a fluid sample, comprising: α-amylase that is substantially calcium-free, a sodium ion, and an α-amylase activity detecting substrate, wherein the composition is substantially free of both chloride ion and a calcium ion source capable of releasing calcium ion in the presence of a chloride ion and α-amylase ~~and wherein the α-amylase is capable of being activated by the sodium ion in proportion to the amount of the chloride ion in the fluid sample~~.

13. (original) A composition as in claim 12 further comprising a compound capable of forming a chelate with a calcium ion and a calcium chelate compound.

14. (original) A composition according to claim 13, wherein said compound capable of forming a chelate with a calcium ion is a member selected from the group consisting of ethylenediaminetetraacetic acid, trans-1,2-cyclohexanediamine-N,N,N',N'-tetraacetic acid, glycol ether diamine tetraacetic acid, iminotetraacetic acid, and diaminopropanetetraacetic acid.

15. (original) A composition according to claim 13, wherein said compound capable of forming a chelate with a calcium ion is ethylenediaminetetraacetic acid.

16. (original) The composition according to claim 13, wherein said calcium chelate compound is calcium-ethylenediaminetetraacetic acid.

17. (original) The composition according to claim 12, wherein said α-amylase activity detecting substrate is a member selected from the group consisting of 2-chloro-4-nitrophenyl-α-D-maltotrioside, 4-nitrophenyl-α-D-maltopentaoside and α-glucosidase, 2-chloro-4-nitrophenyl-β-D-maltopentaoside and α-glucosidase and β-glucosidase, 4-nitrophenyl-α-D-maltoheptaoside, α-glucosidase, and 2-chloro-4-nitrophenyl-β-D-maltoheptaoside and α-glucosidase and β-glucosidase.

18. (original) The composition according to claim 12, wherein said α-amylase activity detecting substrate is 2-chloro-4-nitrophenyl-α-D-maltotrioside.

19. (currently amended) The composition of claim 12, wherein said sodium ion is in the form of sodium citrate.

20. (currently Amended) The composition of claim 12, wherein said sodium ion is in the form of sodium acetate.

21. (withdrawn) A method of activating calcium-free α-amylase for enzymatic activity comprising mixing chloride ion with calcium-free α-amylase in the presence of excess sodium ion.

22. (withdrawn) A method for determining the concentration of sodium ions in samples, comprising:

preparing an enzyme reagent, said enzyme reagent including:

α-amylase that is substantially calcium-free; and
an α-amylase activity detecting substrate; and

combining the enzyme reagent with excess chloride ion, and a sample containing sodium ion to be assayed, the chloride ion being present in a higher concentration than said sodium ion;

assaying the quantity of α-amylase formed due to the presence of sodium ions and

chloride ions in said sample; and determining the quantity of said sodium ions by reference to said assay of α-amylase.

23. (withdrawn) The method of claim 22, wherein a calcium-binding compound is combined with the enzyme reagent, the excess chloride ion, and the sample containing sodium ion to be assayed before the α-amylase quantity is determined.

24. (withdrawn) The method of claim 22, wherein said calcium-binding compound is ethylenediaminetetraacetic acid.

25. (new) The method of claim 1, wherein α-amylase is not substantially activated by calcium ion.